

Dynamics of mitochondrial transport in axons

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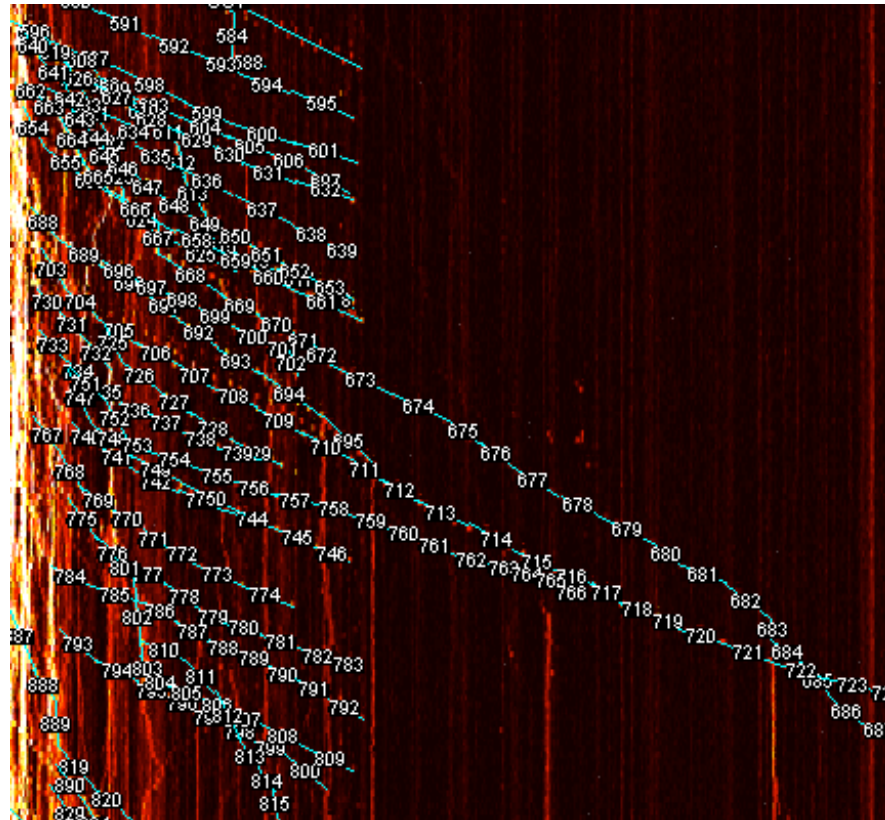
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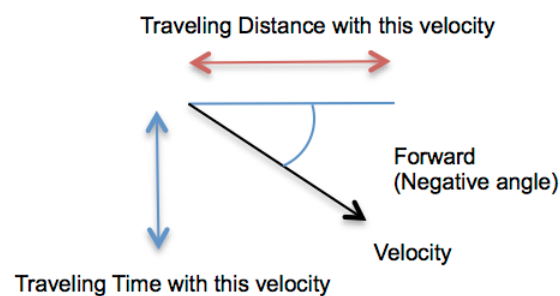
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Methods

Analysis of mitochondrial motility



Velocity distribution of moving mitochondria was analyzed by measuring the velocity of a mitochondrion as it travels through the axon. At first, straight lines are drawn on the kymograph along the trajectories of moving mitochondria using ImageJ. The length of each line is short enough for that to be well fitted in corresponding interval. After that, length and angle of each line are measured in pixels.

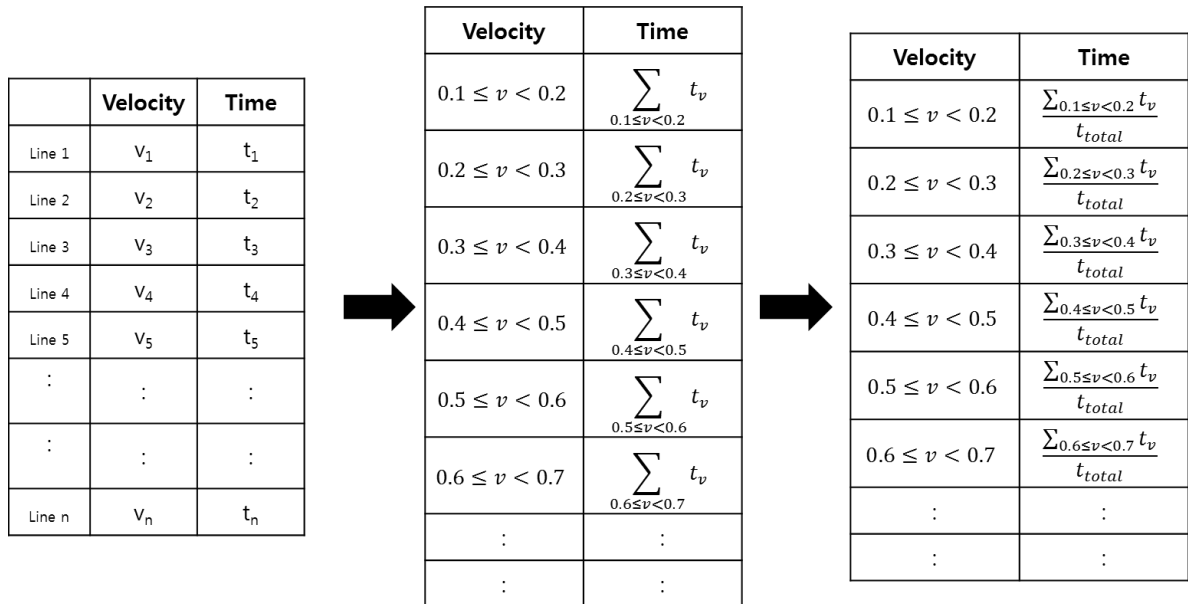


Velocity of each mitochondrion was then calculated from the slope of line with equation below.

$$\text{Velocity} = \left(\cot(-\text{Angle}) \frac{\text{pixel}}{\text{pixel}} \right) \times \left(\frac{\left(\frac{\text{branch length}}{\text{kymograph width}} \right) \frac{\text{um}}{\text{pixel}}}{\left(\frac{\text{time lapse}}{\text{kymograph height}} \right) \frac{\text{s}}{\text{pixel}}} \right) \quad (1.1)$$

Also, traveling time of a mitochondrion with each velocity is calculated for velocity distribution with equation (1.2).

$$\text{Traveling Time} = \text{abs} \left(\text{Length} \times \sin(\text{Angle}) \frac{\text{pixel}}{\text{pixel}} \right) \times \left(\frac{\text{time lapse}}{\text{kymograph height}} \right) \frac{\text{s}}{\text{pixel}} \quad (1.2)$$

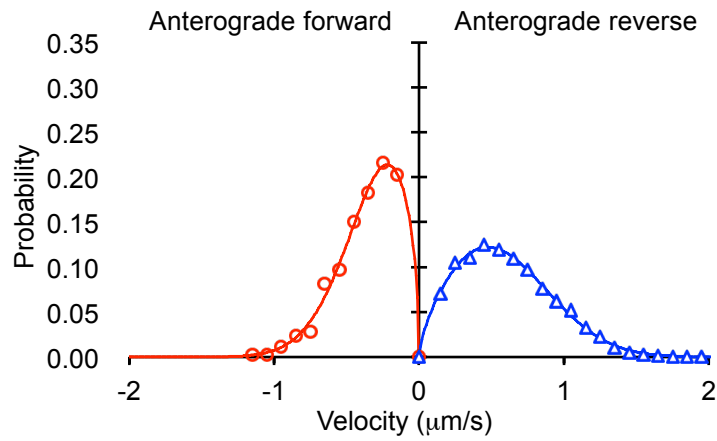


Afterward, binning of all velocity data was done with bin size of 0.1 μm /second followed by normalization of data. Each velocity data was weighted with traveling time of a mitochondrion with that velocity. Note that the velocity from 0 μm /second to 0.1 μm /second was treated as 'stop', thus not included in velocity distribution of moving mitochondria. And the velocities of forward movement and reverse movement were separately normalized.

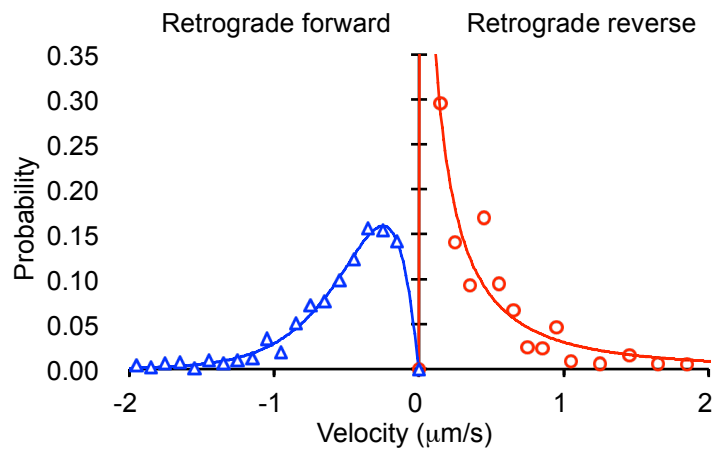
2. Derivation of Fokker-Planck Equation

Normal axon

Anterograde movement



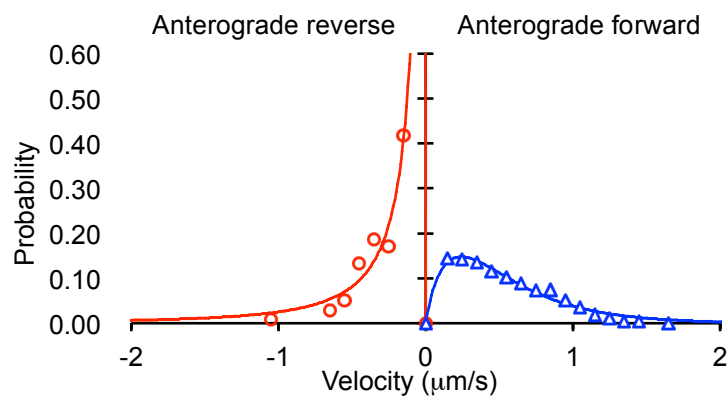
Retrograde movement



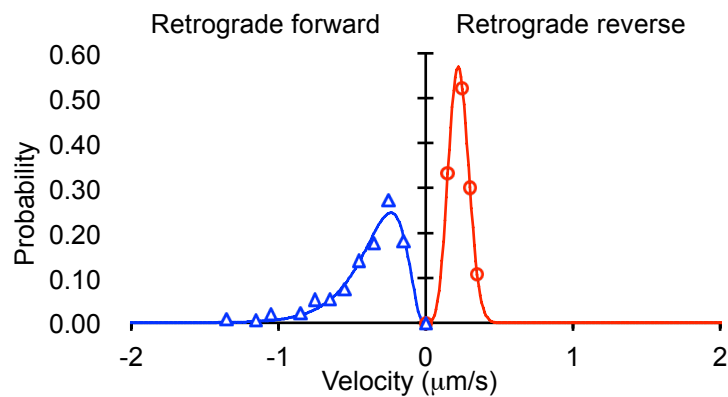
	A	B	n	m
Anterograde forward	0.268	1.612	0.671	-0.645
Anterograde reverse	0.551	4.284	0.460	-0.419
Retrograde forward	2.793	4.584	1.167	1.228
Retrograde reverse	15975.079	13.191	0.928	1.737

Axon overexpressing parkin

Anterograde movement



Retrograde movement



	A	B	n	m
Anterograde forward	1.744	3.819	0.964	1.087
Anterograde reverse	271159.278	16.192	0.783	1.621
Retrograde forward	163.715	10.244	2.551	2.663
Retrograde reverse	706.521	77.217	3.765	2.128

Supplementary Figures

Supplementary Figure 1: Ratio Red/Green image of retrograde movement

kymograph from Figure 1B. The ratiometric image emphasizes that there is little spontaneous photo-activation from the imaging conditions used.